

Figure 1: Space filling representation of the Fv fragment of the antibody 4-4-20

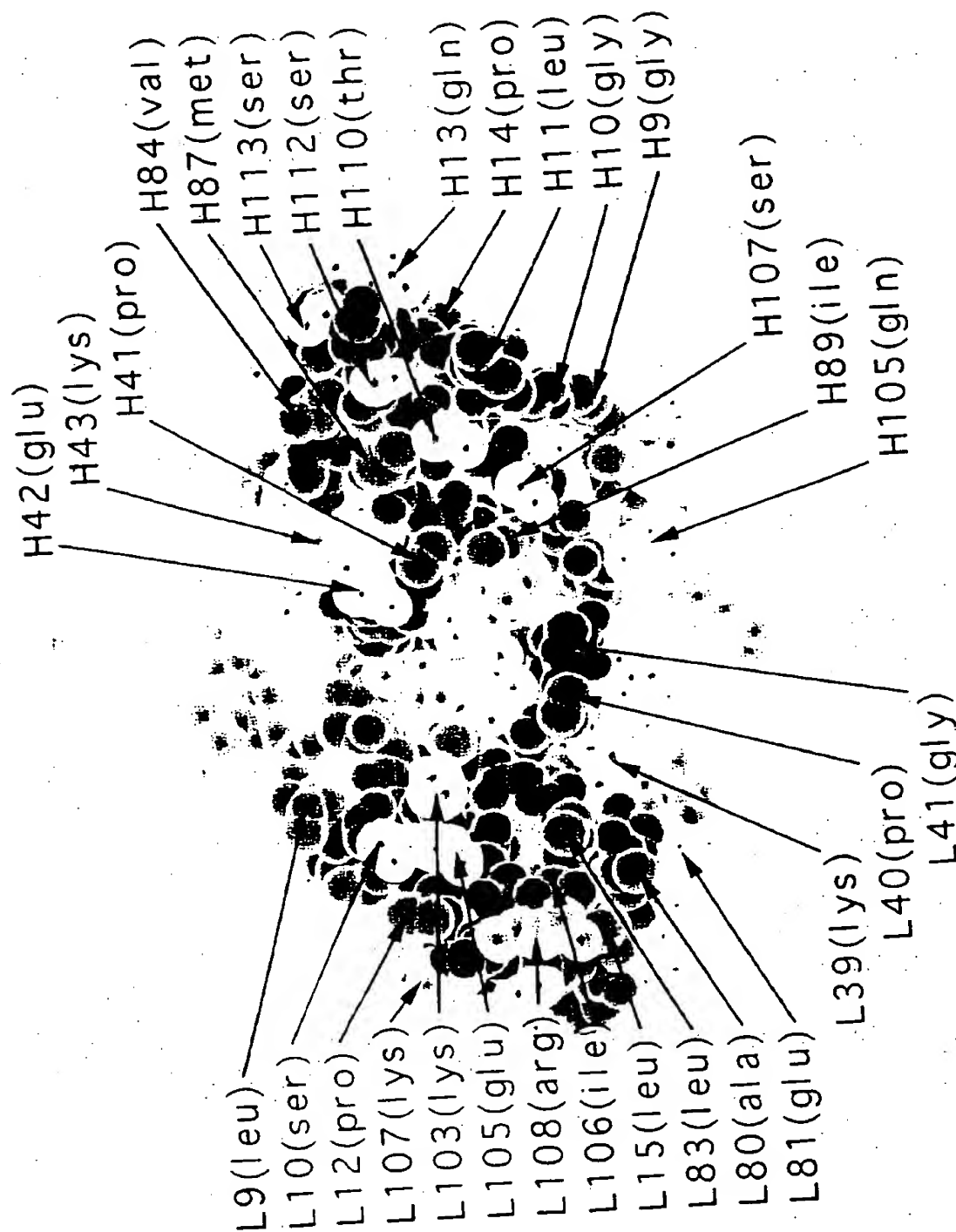


Figure 2a: Variable/constant domain interface residues for VL

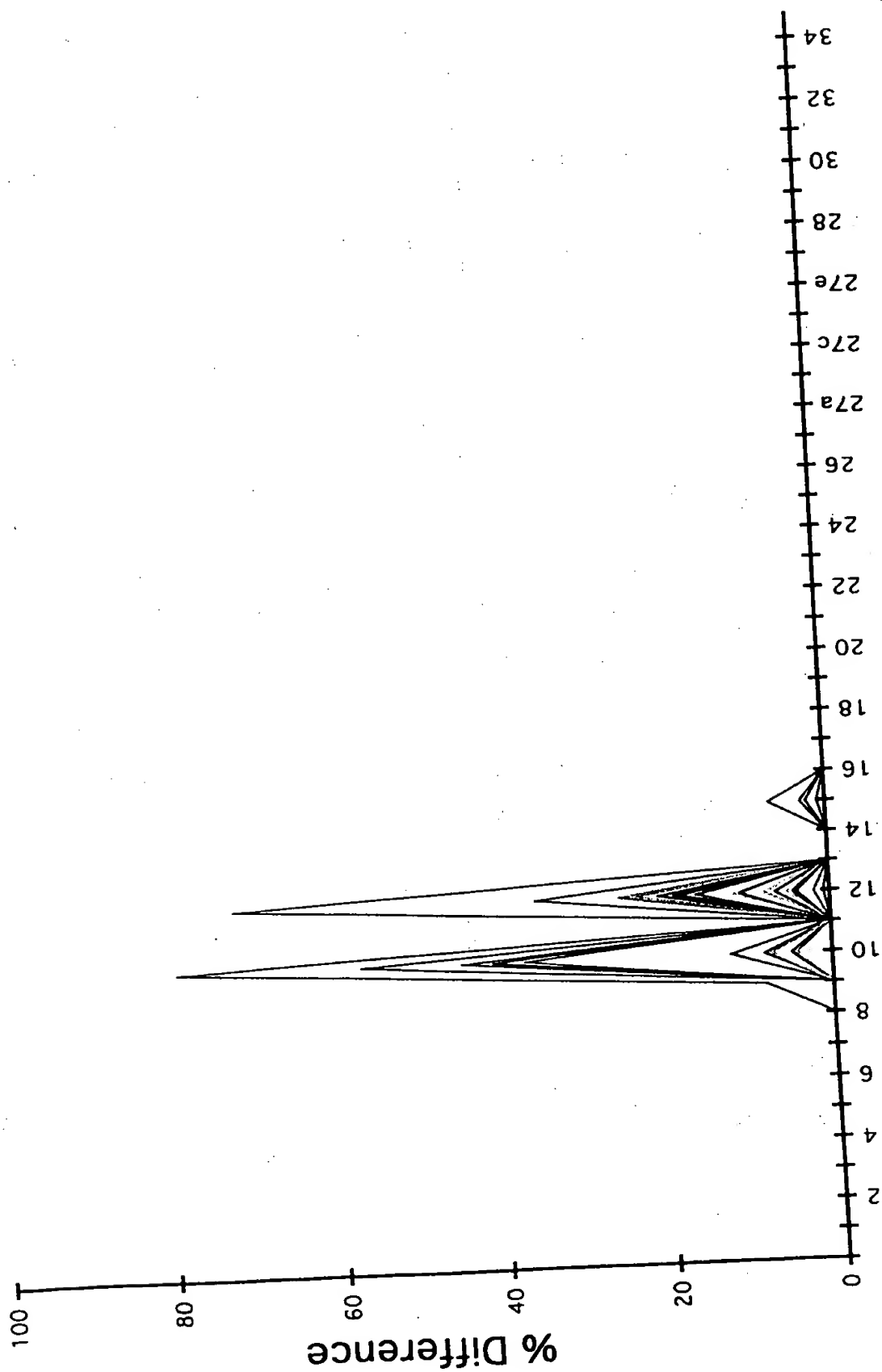


Figure 2a: Variable/constant domain interface residues for VL
(cont.)

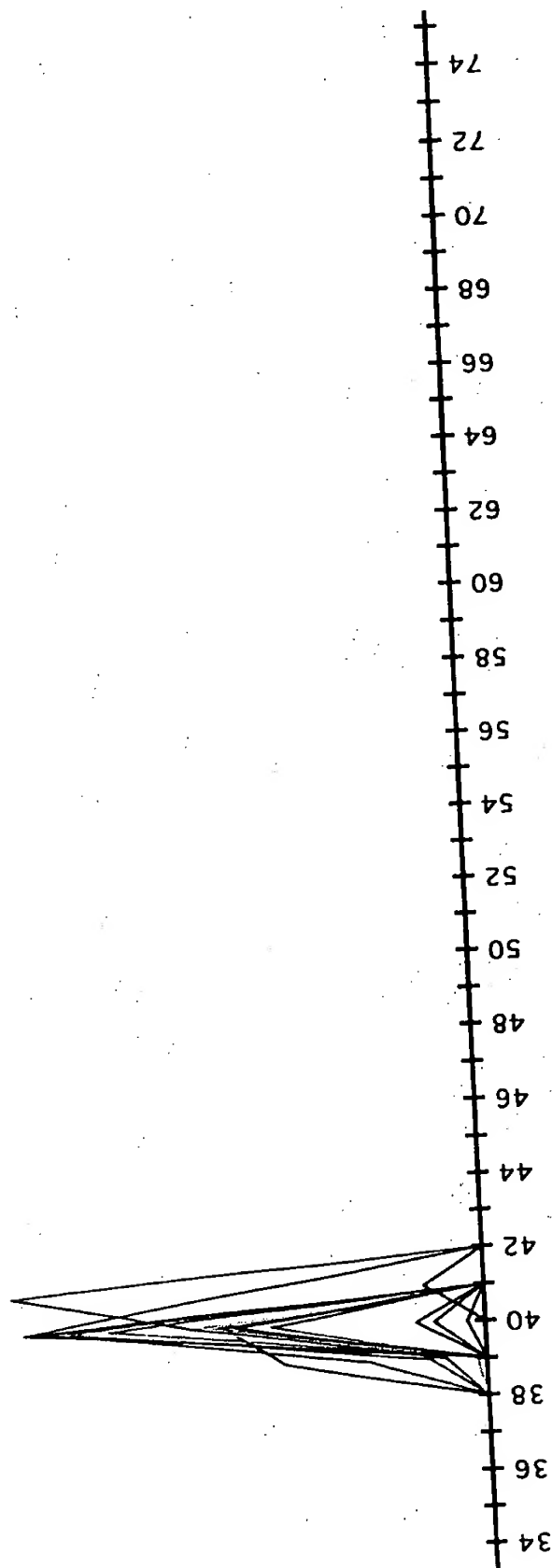


Figure 2a: Variable/constant domain interface residues for VL
(cont.)

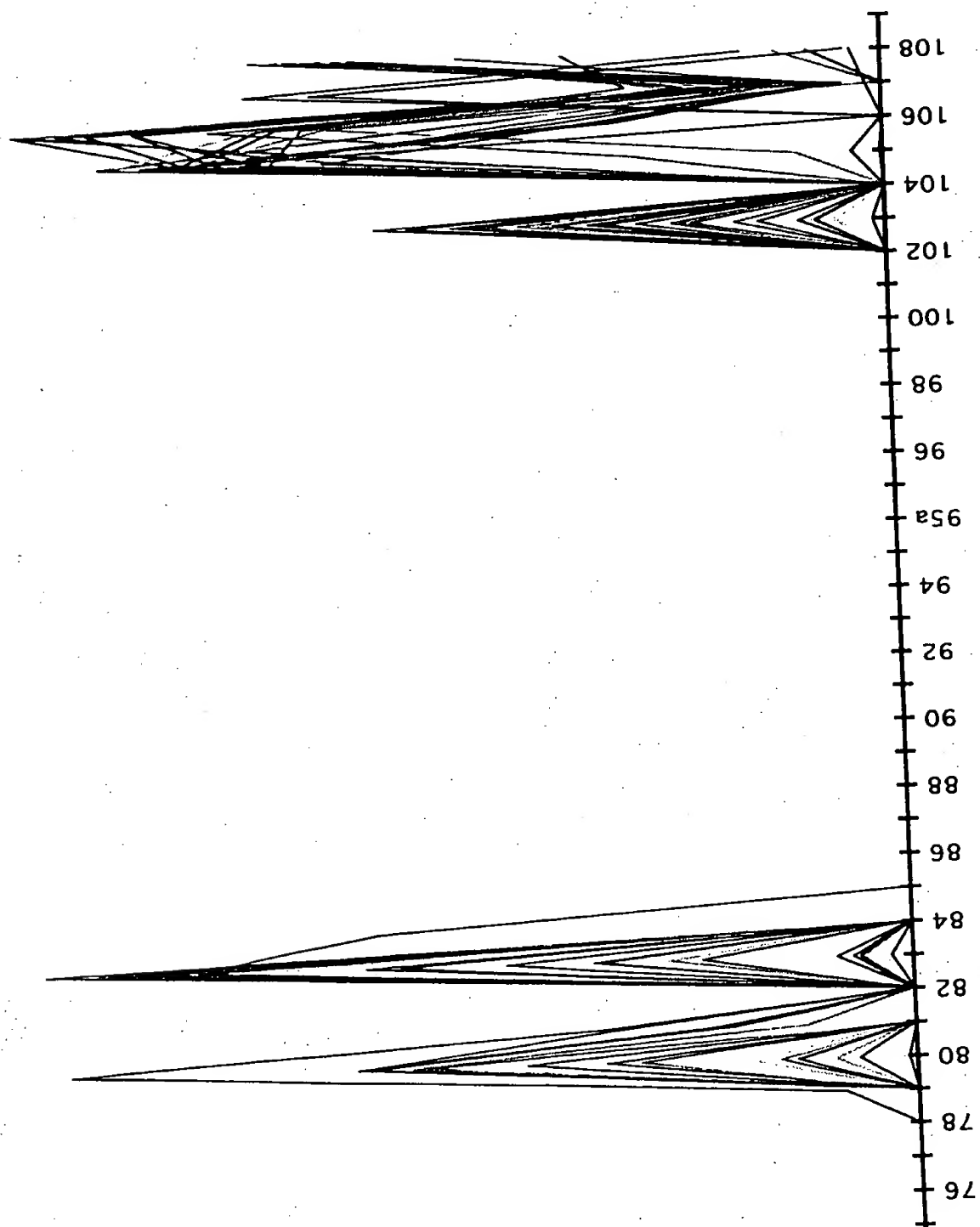


Figure 2a: Variable/constant domain interface residues for VL
(cont.)

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22		
1acy	D	I	V	M	T	Q	S	P	A	S	L	V	●	V	S	L	●	G	Q	R	A	T	I	S
1baf	Q	I	V	L	T	Q	S	P	A	I	●	S	●	A	S	P	G	E	K	V	T	M	T	
1bbj	D	I	Q	M	T	Q	S	P	A	S	L	S	●	V	S	V	G	E	T	V	T	I	T	
1cbv	D	V	V	M	T	Q	T	P	L	S	L	P	●	V	S	L	G	D	Q	A	S	I	S	
1dfb	D	I	Q	M	T	Q	S	P	S	T	L	S	■	A	S	V	G	D	R	V	T	I	T	
1fdl	D	I	Q	M	T	Q	S	P	A	S	L	S	●	A	S	V	G	E	T	V	T	I	T	
1fig	E	N	V	L	T	Q	S	P	A	I	M	S	●	A	S	P	G	E	K	V	T	M	A	
1frg	D	I	V	M	T	Q	S	P	S	S	●	T	■	V	T	A	G	E	K	V	T	M	S	
1fvd	D	I	Q	M	T	Q	S	P	S	S	●	S	■	A	S	V	G	D	R	V	T	I	T	
1ggb	D	I	V	L	T	Q	S	P	G	S	L	A	●	V	S	L	G	Q	R	A	T	I	S	
1gig	Q	A	V	V	T	Q	E	■	S	A	L	T	●	V	T	A	G	E	K	V	T	M	S	
1hin	D	I	V	M	T	Q	S	P	S	S	▲	P	●	V	S	L	G	D	Q	A	S	I	S	
1igi	D	V	V	M	T	Q	T	P	L	S	A	T	■	T	S	P	G	E	T	V	T	L	T	
1ind	■	A	V	V	T	Q	E	■	S	S	L	P	■	V	S	L	G	D	Q	A	S	I	S	
1jel	D	V	L	M	T	Q	T	P	L	S	L	P	■	A	S	L	G	D	R	V	T	I	S	
1mam	D	I	Q	M	T	Q	T	T	S	S	L	S	■	G	S	L	G	Q	S	V	T	I	S	
1mco	P	S	A	L	T	Q	P	■	P	S	A	S	■	T	S	V	G	D	R	V	T	I	T	
1nca	D	I	V	M	T	Q	S	P	K	F	M	S	●	V	S	L	G	D	Q	A	S	I	S	
2cgr	E	L	V	M	T	Q	S	P	L	S	L	P	●	V	N	L	G	D	Q	A	S	I	S	
2dbl	D	V	V	M	T	Q	I	P	L	●	L	P	●	V	S	L	G	D	Q	A	S	I	S	
2f19	D	I	Q	M	T	Q	T	T	S	S	L	S	●	A	S	L	G	D	R	V	T	I	S	
2fb4	Q	S	V	L	T	Q	P	■	P	S	A	S	●	G	T	P	G	Q	R	V	T	I	T	
2fbj	E	I	V	L	T	Q	S	P	A	I	T	A	■	A	S	P	G	E	K	V	T	M	T	
2hfl	D	I	V	L	T	Q	S	P	A	I	●	S	●	V	S	L	G	D	Q	A	S	I	S	
2igf	D	V	L	M	T	Q	T	P	L	S	L	P	●	V	S	A	G	E	R	V	T	M	S	
2mcp	D	I	V	M	T	Q	S	P	S	S	L	S	■	V	T	P	G	N	S	V	S	L	S	
3hfm	D	I	V	L	T	Q	S	P	A	T	L	S	●	V	S	L	G	D	Q	A	S	I	S	
4fab	D	V	V	M	T	Q	T	P	L	S	■	P	●	V	S	L	G	D	R	V	S	I	S	
6fab	D	I	Q	M	T	Q	I	P	S	S	L	S	■	A	S	L	G	Q	T	A	R	I	T	
8fab	■	E	L	T	Q	P	■	P	P	S	▲	V	▼	V	S	P	G	Q	T	A	R	I	T	

Figure 2a: Variable/constant domain interface residues for VL

23	24	25	26	27	27a	27b	27c	27d	27e	27f	28	29	30	31	32	33	34	35	36	37	38	39	O	
C	R	A	S	E	S	V	D		S	Y	G	K	S	F	M	H	W	Y	Q	Q	K	■		
C	S	A	S	S								S	V	Y	Y	M	Y	W	Y	Q	Q	K		
C	R	A	S	E					N	I	Y	S	N	L	A	W	Y	Y	Q	Q	K			
C	R	S	S	Q	S	L	V		H	S	N	G	N	T	Y	L	H	W	Y	L	Q	K		
C	R	A	S	Q					S	I	S	R	W	L	A	W	Y	Y	Q	Q	K			
C	R	A	S	Q					N	I	H	N	Y	L	A	W	Y	Y	Q	Q	K			
C	R	A	S	G					V	S	S	T	Y	L	H	W	Y	Y	Q	Q	K			
C	R	A	S	S	S				K	R	K	N	F	L	T	W	Y	Y	H	Q	K			
C	K	S	S	Q	S	L	F	N	S	G	D	V	N	T	A	V	A	W	Y	Q	Q	K		
C	R	A	S	Q					D	D	G	N	S	F	L	H	W	Y	Y	Q	Q	K		
C	R	A	S	E	S	V	D		V	T	T	S	N	Y	A	N	W	Y	Y	Q	E	K		
C	R	S	S	T	G	A			S	G	K	Q	K	N	Y	L	T	W	Y	Q	Q	K		
C	T	S	S	Q	S	L	F	N	S	H	S	N	G	N	T	Y	L	N	W	Y	L	Q	K	
C	R	S	S	Q	S	L	V		V	T	T	S	N	Y	A	N	W	Y	Y	Q	E	K		
C	R	S	S	T	G	A			H	G	N	G	N	T	Y	L	E	W	Y	L	Q	K		
C	R	S	S	Q	S	I	V				D	I	Y	N	Y	L	N	W	Y	Q	Q	K		
C	R	A	S	Q					V	G	G	Y	N	Y	V	S	W	Y	Y	Q	Q	H		
C	T	G	T	S	S	D					D	V	S	T	A	V	V	W	Y	Q	Q	K		
C	K	A	S	Q					H	S	N	G	N	T	Y	L	H	W	Y	L	Q	K		
C	R	P	S	Q	S	L	V		H	S	N	G	N	T	Y	L	H	W	Y	L	Q	K		
C	R	S	S	Q	S	L	I				D	I	S	N	Y	L	N	W	Y	Q	Q	K		
C	R	A	S	Q					N	I	G	S	S	T	V	N	W	Y	Y	Q	Q	L		
C	S	G	T	S	S						S	V	S	S	L	N	W	Y	Y	Q	Q	K		
C	S	A	S	S							S	V	N	Y	M	Y	W	Y	Y	Q	Q	K		
C	S	A	S	S							L	S	D	G	D	T	Y	L	E	W	Y	L	Q	K
C	R	S	N	Q	T	I	L				S	G	N	Q	K	N	F	L	A	W	Y	Q	Q	K
C	K	S	S	Q	S	L	L	N	S		S	I	G	N	N	L	H	W	Y	Y	Q	Q	K	
C	R	A	S	Q					H	S	Q	G	N	T	Y	L	R	W	Y	L	Q	K		
C	R	S	S	Q	S	L	V				D	I	N	N	F	L	N	W	Y	Y	Q	Q	K	
C	S	A	N	A							L	P	N	Q	Y	A	Y	W	Y	Y	Q	Q	K	

Figure 2a: Variable/constant domain interface residues for VL
(cont.)

	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	
P	■	G	Q	P	P	K	V	L	I	Y	I	A	S	N	L	E	S	G	V	P	A	R	F	
P		G	S	S	P	R	L	L	I	Y	D	T	S	N	L	A	S	G	V	P	V	R	F	
Q	■	G	K	S	P	Q	L	L	V	Y	A	A	T	N	L	A	D	G	V	P	S	R	F	
P		G	Q	S	P	K	L	L	I	Y	K	V	S	N	R	F	S	G	V	P	D	R	F	
P	■	G	K	V	P	K	L	L	I	Y	K	A	S	S	L	E	S	G	V	P	S	R	F	
Q	●	G	K	S	P	Q	L	L	V	Y	Y	T	T	T	L	A	D	G	V	P	S	R	F	
S	▲	G	A	S	P	K	L	L	I	Y	S	T	S	N	L	A	S	G	V	P	A	R	F	
P		G	Q	P	P	K	L	L	I	Y	W	A	S	T	R	E	S	G	V	P	D	R	F	
P	■	G	K	A	P	K	L	L	I	Y	S	A	S	F	L	E	S	G	V	P	S	R	F	
P	■	G	Q	P	P	K	L	L	I	Y	R	S	S	N	L	I	S	G	I	P	D	R	F	
P	■	G	Q	P	P	K	L	L	I	G	G	T	N	N	R	A	P	G	V	P	A	R	F	
P	▲	D	H	L	F	T	G	L	I	Y	W	A	S	T	R	E	S	G	V	P	D	R	F	
P		G	Q	P	P	K	V	L	I	Y	K	V	S	N	R	F	S	G	V	P	D	R	F	
A		G	Q	S	P	K	L	L	I	Y	G	T	N	N	R	A	P	G	V	P	A	R	F	
P	■	D	H	L	F	T	G	L	I	Y	S	I	S	S	R	F	S	G	V	P	D	R	F	
P	■	G	Q	S	P	K	L	L	I	Y	Y	T	S	R	L	H	S	G	V	P	S	R	F	
P	■	D	G	T	V	K	L	L	I	Y	E	V	N	K	R	P	S	G	V	P	D	R	F	
A	■	G	K	A	P	K	V	L	I	Y	W	A	S	T	R	H	I	G	V	P	D	R	F	
P	■	G	Q	S	P	K	L	L	I	Y	R	V	S	N	R	F	S	G	V	P	D	R	F	
P	■	G	Q	S	P	K	L	L	M	Y	K	V	S	N	R	F	Y	G	V	P	D	R	F	
P		G	Q	S	P	K	L	L	I	Y	Y	T	S	R	L	H	S	G	V	P	S	R	F	
P	●	D	G	T	V	K	L	L	I	Y	R	D	A	M	R	P	S	G	V	P	D	R	F	
P		G	M	A	P	K	L	L	I	Y	R	E	I	S	K	L	A	S	G	V	P	A	R	F
S	▲	G	T	S	P	K	P	W	I	Y	E	D	T	S	K	L	A	S	G	V	P	V	R	F
S	■	G	T	S	P	K	R	W	I	Y	K	V	S	N	R	F	S	G	V	P	D	R	F	
P	■	G	Q	S	P	K	L	L	I	Y	G	A	S	T	R	E	S	G	V	P	D	R	F	
P	▲	G	Q	P	P	K	L	L	I	Y	K	Y	A	S	Q	S	I	S	G	I	P	S	R	F
S	▲	H	E	S	P	R	L	L	I	Y	K	V	S	N	R	F	S	G	V	P	D	R	F	
P		G	Q	S	P	K	V	L	I	Y	K	V	S	N	R	S	Q	S	G	V	P	S	R	F
P	■	D	G	T	I	K	L	L	I	Y	F	T	S	R	S	P	S	G	V	P	S	R	F	
P		G	R	A	P	V	M	V	I	Y	K	D	T	Q	R	P	S	G	I	P	Q	R	F	
P		G																						

Figure 2a: Variable/constant domain interface residues for VL
(cont.)

	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83				
S	G	S	G	S	G	S	R	T	D	F	T	L	T	I	D	P	V	E	A	▼	D	■	D	A	◆
S	G	S	G	S	G	S	G	T	S	Y	S	L	T	I	S	R	M	E	A		E		D	A	●
S	G	S	G	S	G	S	G	T	Q	Y	S	L	K	I	N	S	L	Q	S	▼	E	■	D	F	◆
S	G	S	G	S	G	S	G	T	D	F	T	L	K	I	S	R	V	E	A		E		D	L	●
S	G	S	G	S	G	S	G	T	E	F	T	L	T	I	S	S	L	Q	P		D		D	F	
S	G	S	G	S	G	S	G	T	Q	Y	S	L	K	I	N	S	L	Q	P		E		D	F	
S	G	S	G	S	G	S	G	T	S	Y	S	L	T	I	S	S	V	E	●	◆	E	■	D	A	◆
S	G	S	G	S	G	S	G	T	D	F	T	L	T	I	T	S	V	Q	A		E		D	L	●
S	G	S	G	S	G	S	G	T	D	F	T	L	T	I	S	S	L	Q	P	●	E		D	F	▼
S	G	S	G	S	R	S	G	T	D	F	T	L	T	I	S	P	V	E	A		D		D	V	■
S	G	S	G	S	G	S	G	T	D	K	A	L	T	I	T	G	A	Q	T	■	E		D	E	▲
T	G	S	G	S	G	S	G	T	D	F	T	L	T	I	S	S	V	Q	A		E		D	L	●
S	G	S	G	S	G	S	G	T	D	F	T	L	K	I	S	R	V	E	A		E		D	L	
S	G	S	G	S	L	I	G	D	K	A	A	L	T	I	T	G	A	Q	T		E		D	E	▼
S	G	S	G	S	G	S	G	T	D	K	A	L	T	I	S	R	V	Q	A	●	E		D	L	▼
S	G	S	G	S	G	S	G	T	D	F	T	L	K	I	S	N	L	N	Q	●	E		D	M	◆
S	G	S	G	S	K	S	G	T	D	Y	S	L	T	I	S	G	L	Q	A		E	■	D	L	◆
A	G	S	G	S	G	S	G	T	D	Y	T	L	T	I	S	S	V	Q	A	▲	E		D	L	◆
S	G	S	G	S	G	S	G	T	A	F	T	L	K	I	S	R	V	E	A		E		D	L	
S	G	S	G	S	G	S	G	T	D	F	T	L	K	I	S	R	V	E	A		E		D	I	
S	G	S	G	S	G	S	G	T	D	Y	S	L	T	I	S	N	L	E	H	●	E		D	E	■
S	G	S	G	S	K	S	G	T	A	S	L	A	I	G	G	L	Q	E	S	●	E		D	E	◆
S	G	S	G	S	G	S	G	T	S	Y	S	L	T	I	N	T	M	E	A	▲	E		D	A	◆
S	G	S	G	S	G	S	G	T	S	Y	S	L	T	I	S	S	M	E	T		E		D	A	■
S	G	S	G	S	G	S	G	T	S	Y	S	L	T	I	S	R	V	E	A	■	E		D	L	◆
S	G	S	G	S	G	S	G	T	D	F	T	L	K	I	S	R	V	E	A		E		D	L	◆
S	G	S	G	S	G	S	G	T	D	F	T	L	T	I	S	S	V	Q	A		E	■	D	L	◆
T	G	S	G	S	G	S	G	T	D	F	T	L	T	I	S	S	V	E	T	●	E		D	F	◆
S	G	S	G	S	G	S	G	T	D	F	T	L	S	I	N	S	V	E	A		E		D	L	▲
S	G	S	G	S	G	S	G	T	D	F	T	L	K	I	S	R	V	E	A	▲	E		D	I	■
S	G	S	G	S	G	S	G	T	D	Y	S	L	T	I	S	N	L	E	Q		E		D	E	■
S	S	S	S	T	S	S	G	T	T	V	T	L	T	I	S	G	V	Q	A		E		D	E	■

Figure 2a: Variable/constant domain interface residues for VL
(cont.)

84	85	86	87	88	89	90	91	92	93	94	95	95a	95b	96	97	98	99	100	101	102	103	O
A	T	Y	Y	C	Q	Q	Q	N	N		E	D	P	P	T	F	G	A	G	T	K	■
A	T	Y	Y	C	Q	Q	Q	W	S	S	Y	P	P	I	T	F	G	V	G	T	K	■
G	S	Y	Y	C	Q	H	F	W			G	T	P	Y	T	F	G	G	G	T	R	
G	V	Y	F	C	S	Q	S	T			H	V	P	L	T	F	G	A	G	T	K	■
A	T	Y	Y	C	Q	Q	Y					N	S	Y	S	F	G	P	G	T	K	■
G	S	Y	Y	C	Q	H	F	W			S	T	P	R	T	F	G	G	G	T	K	
A	T	Y	Y	C	Q	Q	Y	S			G	Y	P	L	T	F	G	A	G	T	K	
A	I	Y	Y	C	Q	N	D	Y			S	H	P	L	T	F	G	A	G	T	K	▲
A	T	Y	Y	C	Q	Q	H	Y			T	T	P	P	T	F	G	Q	G	T	K	▲
A	T	Y	Y	C	Q	Q	S	N			E	D	P	L	T	F	G	A	G	T	K	
A	T	Y	Y	C	Q	Q	S	N			S	N	H	W	V	F	G	G	G	T	K	
A	I	Y	F	C	A	L	W	Y			S	N	P	L	T	F	G	G	G	T	K	■
A	V	Y	Y	C	Q	N	D	Y			S	N	P	P	T	F	G	G	G	T	K	▲
G	I	Y	F	C	S	Q	T	T			H	V	P	P	T	F	G	G	G	T	K	▲
A	R	Y	F	C	A	L	W	Y			S	N	L	W	V	F	G	G	G	T	K	▲
A	V	Y	Y	C	F	Q	G	S			H	V	P	Y	T	F	G	G	G	T	K	▲
A	T	Y	I	C	Q	Q	G	N			T	L	P	F	T	F	G	S	G	T	K	▲
A	D	Y	Y	C	S	S	Y	E			S	D	N	F	V	F	G	T	G	T	K	
A	L	Y	Y	C	Q	Q	H	Y			S	P	P	W	T	F	G	G	G	T	K	■
G	V	Y	F	C	S	Q	G	T			H	V	P	Y	T	F	G	G	G	T	K	■
G	I	Y	F	C	S	Q	S	S			H	V	P	P	T	F	G	G	G	T	K	■
A	T	Y	F	C	Q	Q	Q	G	S		T	L	P	P	T	F	G	G	G	T	K	■
A	D	Y	Y	C	A	A	W	D		V	S	L	N	A	Y	V	F	G	T	G	K	
A	I	Y	Y	C	Q	Q	W	T			Y	P	L	I	T	F	G	A	G	T	K	●
A	E	Y	Y	C	Q	Q	W				G	R	N	P	T	F	G	G	G	T	K	■
A	V	Y	Y	C	F	Q	G	S			H	V	P	P	T	F	G	G	G	T	K	▲
A	V	Y	Y	C	Q	N	D	H			S	Y	P	L	T	F	G	A	G	T	K	
A	M	Y	F	C	Q	Q	S	N			S	W	P	Y	T	F	G	G	G	T	K	▲
G	V	Y	F	C	S	Q	S	T			H	V	P	P	T	F	G	G	G	T	K	▲
A	T	Y	F	C	Q	Q	G	N			A	L	P	P	T	F	G	G	G	T	K	
A	D	Y	Y	C	Q	A	W	D			N	S	A	S	I	F	G	G	G	T	K	■

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Figure 2b: Variable/constant domain interface residues for VH

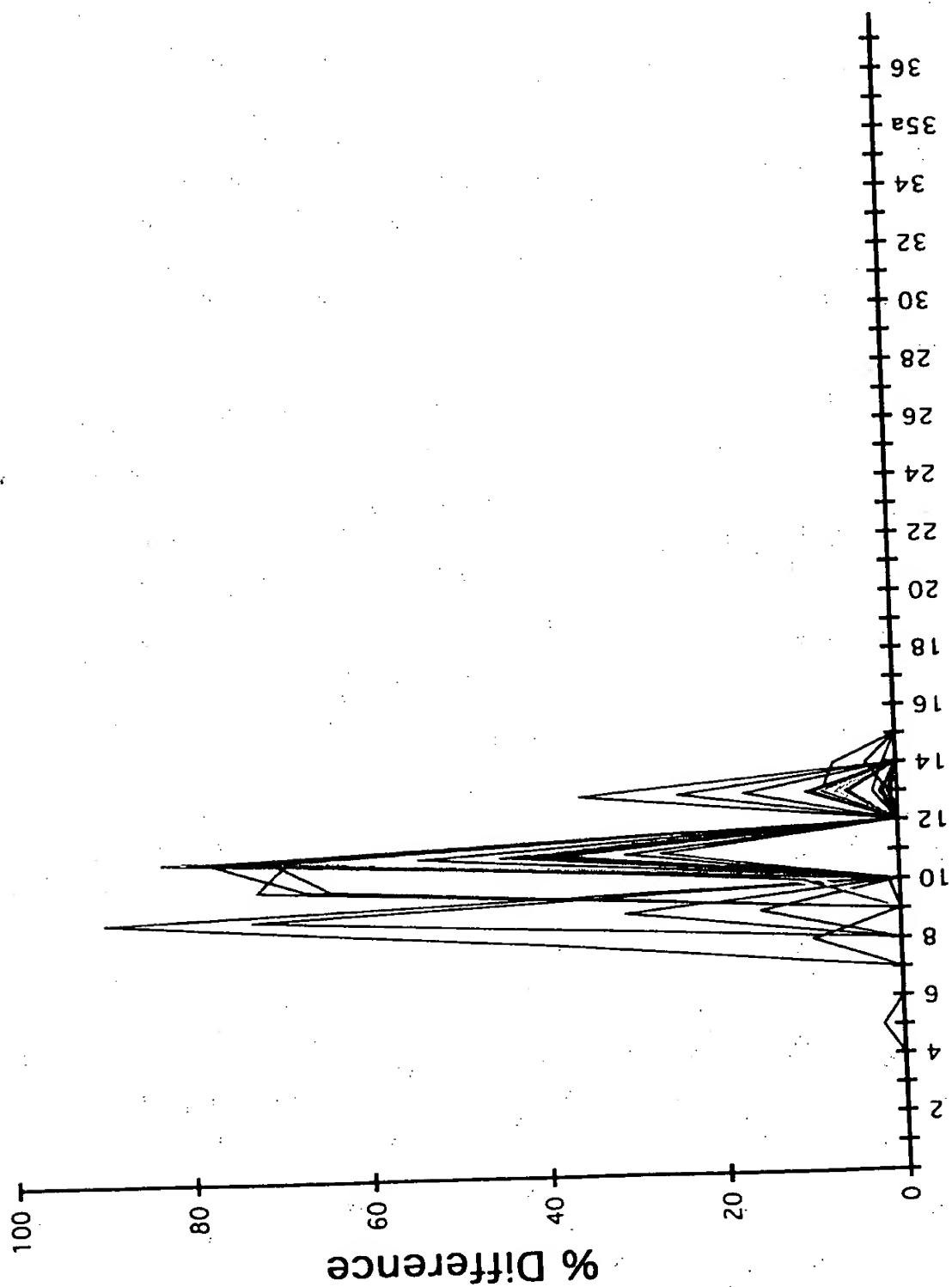


Figure 2b: Variable/constant domain interface residues for VH
(cont.)

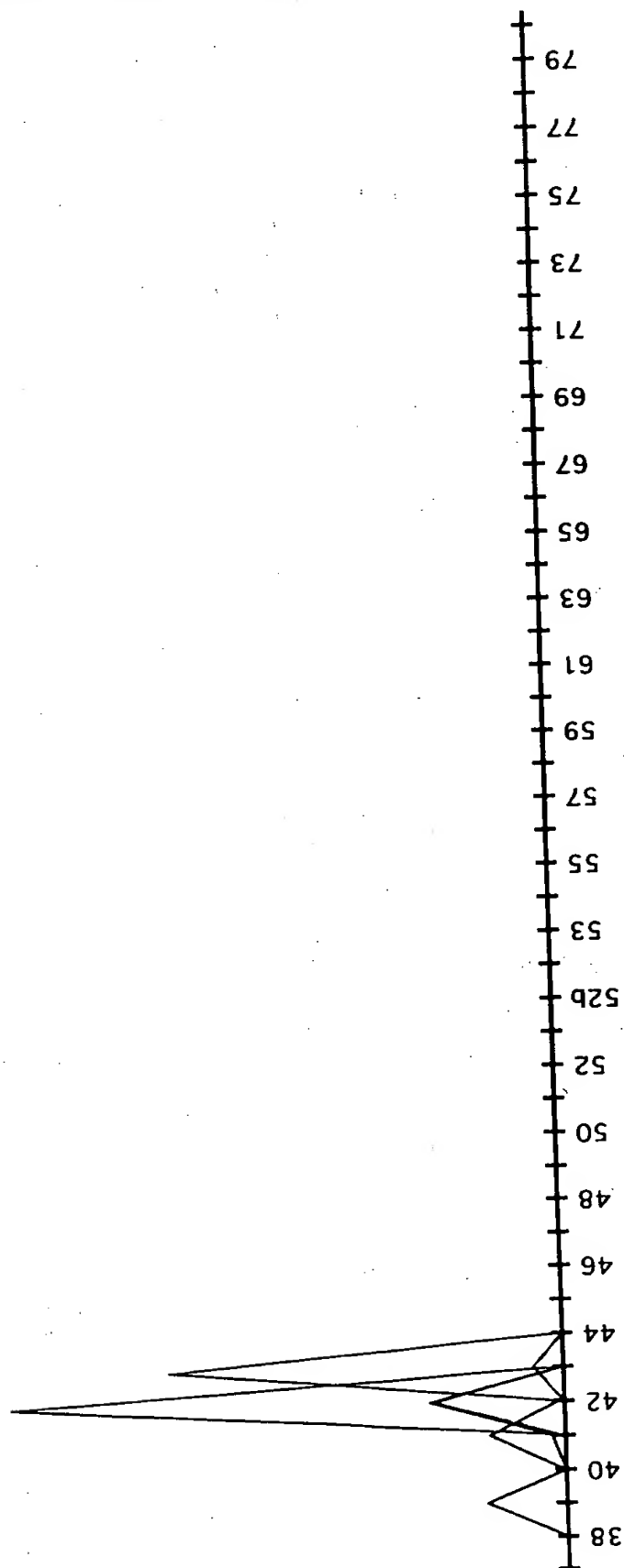


Figure 2b: Variable/constant domain interface residues for VH
(cont.)

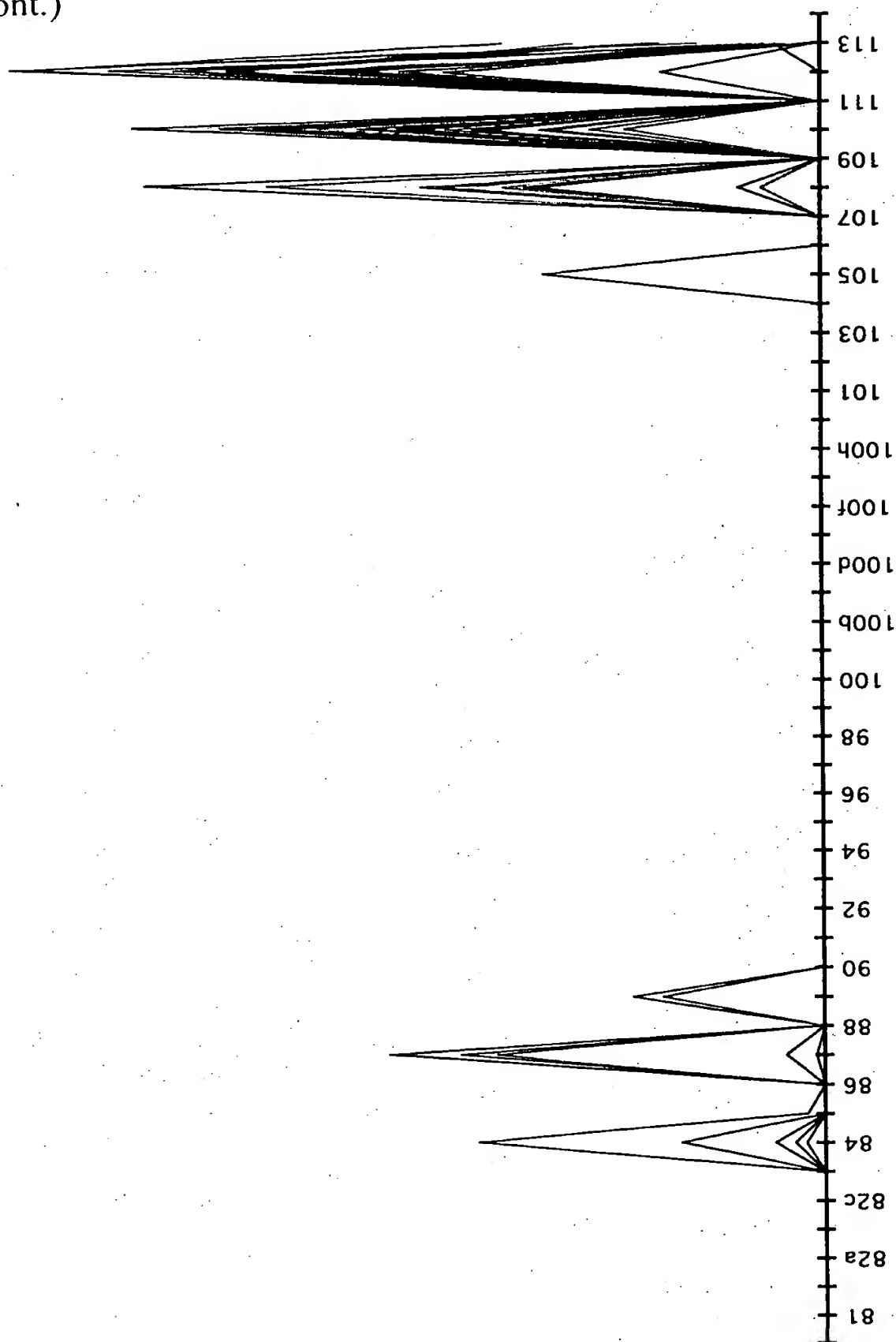


Figure 2b: Variable/constant domain interface residues for VH
(cont.)

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22
1acy	Q	V	K	L	Q	E	S	G	P	A	V	I	K	P	S	Q	S	L	S	L	T	C
1baf	D	V	Q	L	Q	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1bbj		V	Q	L	Q	E	S	G	P	E	L	V	K	P	S	Q	S	L	S	L	T	C
1cbv	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1dfb	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1fdl	Q	V	Q	L	Q	E	S	G	P	D	L	V	K	P	S	Q	S	L	S	L	T	C
1fig	D	V	Q	L	Q	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1frg	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1fvd	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1ggb	Q	V	Q	L	Q	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1gig	Q	V	Q	L	Q	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1hin	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1igi	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1ind	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1jel	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1mam	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1mco	E	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
1nca	P	V	Q	L	V	E	S	G	P	G	L	V	K	P	S	Q	S	L	S	L	T	C
2cgr	Q	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2dbl	Q	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2f19	Q	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2fb4	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2fbj	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2hfl	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2igf	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
2mcp	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
3hfm	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
4fab	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
6fab	E	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C
8fab	A	V	Q	L	V	E	S	G	P	A	L	V	K	P	S	Q	S	L	S	L	T	C

Figure 2b: Variable/constant domain interface residues for VH
(cont.)

23	I	T	K	A	A	T	K	A	A	S	T	A	K	A	K	A	T	K	K	K	K	S	A	K	A	A	S	V	K	I	
24	V	V	A	A	A	V	A	A	A	F	V	A	S	A	G	T	V	A	A	A	A	S	A	A	A	T	V	A	A	A	
25	S	T	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	T	S	S	S	S	S	S	S	S	S	T	S	S	
26	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	
27	F	Y	Y	F	F	F	F	F	F	F	F	F	Y	F	Y	F	D	Y	Y	Y	Y	F	F	Y	F	F	D	F	Y	F	
28	S	S	T	S	T	S	S	T	N	S	L	S	I	T	T	T	S	T	T	A	T	I	D	T	T	T	S	T	T	T	
29	I	I	F	F	F	L	L	F	I	L	L	F	F	L	F	F	I	F	F	F	F	F	F	F	F	F	I	F	F	F	
30	T	T	T	N	N	T	P	S	K	S	I	S	T	S	T	T	N	T	S	T	T	S	S	S	S	S	T	S	T	S	
31	R	T	S																												
32	T	S																													
33	N	D	D	T	D	G	G	S	D	G	S	S	D	G	T	D	L	N	E	N	S	S	K	D	R	D	S	D	S	N	
34	Y	Y	H	N	Y	Y	H	F	T	M	N	Y	F	E	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	C	F	D	Y	N	Y
35	C	A	A	A	A	G	N	G	Y	G	G	G	Y	T	A	Y	Y	G	W	G	G	A	W	W	A	Y	Y	W	G	G	
35a	W	W	I	M	M	V	I	M	I	V	V	M	M	M	M	M	W	M	I	V	V	M	M	I	M	M	W	M	I	M	
35b	H	N	H	N	H	N	N	S	H	S	H	S	N	S	H	S	S	N	E	N	N	Y	S	E	S	E	S	N	N	H	
36	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	
37	I	I	A	V	V	V	I	V	V	I	V	V	V	V	V	V	I	V	V	V	V	V	V	V	V	V	I	V	V	V	
38	R	R	K	R	R	R	V	R	R	R	R	R	R	R	R	K	R	R	K	K	K	K	R	R	K	R	R	R	R	K	R
39	Q	Q	Q	Q	Q	Q	H	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	E	E	Q	Q	Q	Q	Q	Q	K	Q	Q	Q		
40	A	F	K	A	A	P	R	T	A	P	P	T	S	T	S	P	P	A	R	A	R	A	A	R	T	P	F	S	R	A	
41	P	P	P	P	P	P	N	P	P	S	P	P	H	P	H	P	P	P	P	P	P	P	P	P	P	P	P	P	P	P	
42	G	G	E	G	G	G	G	D	G	G	G	D	G	E	A	G	G	G	G	G	G	G	G	G	E	G	G	E	G	G	
43	K	N	Q	K	K	K	K	K	K	K	K	K	K	K	K	K	K	K	H	K	Q	K	K	H	K	K	N	K	Q	K	

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Figure 2b: Variable/constant domain interface residues for VH
(cont.)

100f	E	T		T	T		N	E	F		Y	E	K		E		G		N	V	C	Y		P	T		S	A				
100g																																
100h	Y	P	Y	W	A	R	G	G	A	G	A	G	A	R	Y	P	V	L	S	Y	Y	G	Y	D	Y	Y	D	G	K	S		
100i	F	L	Y	F	F	L	F	F	M	Y	M	F	M	F	F	A	V	S	M	F	F	P	N	F	F	F	G	M	F	F		
101	D	A	G	A	D	D	T	A	D	I	D	A	D	V	D	A	N	D	D	D	D	D	A	D	D	D	D	D	D	D		
102	V	Y	H	Y	I	Y	Y	Y	V	Y	Y	Y	Y	H	V	Y	P	Y	Y	V	S	Y	Y	G	Y	V	Y	Y	Y	Y		
103	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W	W		
104	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G		
105	Q	Q	Q	Q	Q	Q	R	Q	Q	Q	Q	H	H	A	Q	Q	Q	Q	A	Q	Q	Q	Q	Q	A	Q	Q	Q	Q	Q		
106	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G	G		
107	T	T	T	T	T	T	T	T	T	T	T	A	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	V		
108	T	Q	T	L	M	T	L	L	L	S	S	L	S	L	T	L	L	T	S	T	T	P	L	T	T	T	L	S	T	L		
109	V	V	L	V	V	L	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	L	V	V	L	L	V	V	V	L	V
110	T	S	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T	T		
111	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	V	
112	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S	
113	S	E	S	A	S	S	A	A	S	S	S	A	S	A	S	A	S	S	A	S	S	S	A	S	S	S	A	S	S	S	S	

Figure 3: Western blots showing the insoluble (i) and soluble (s) fractions of cell extracts

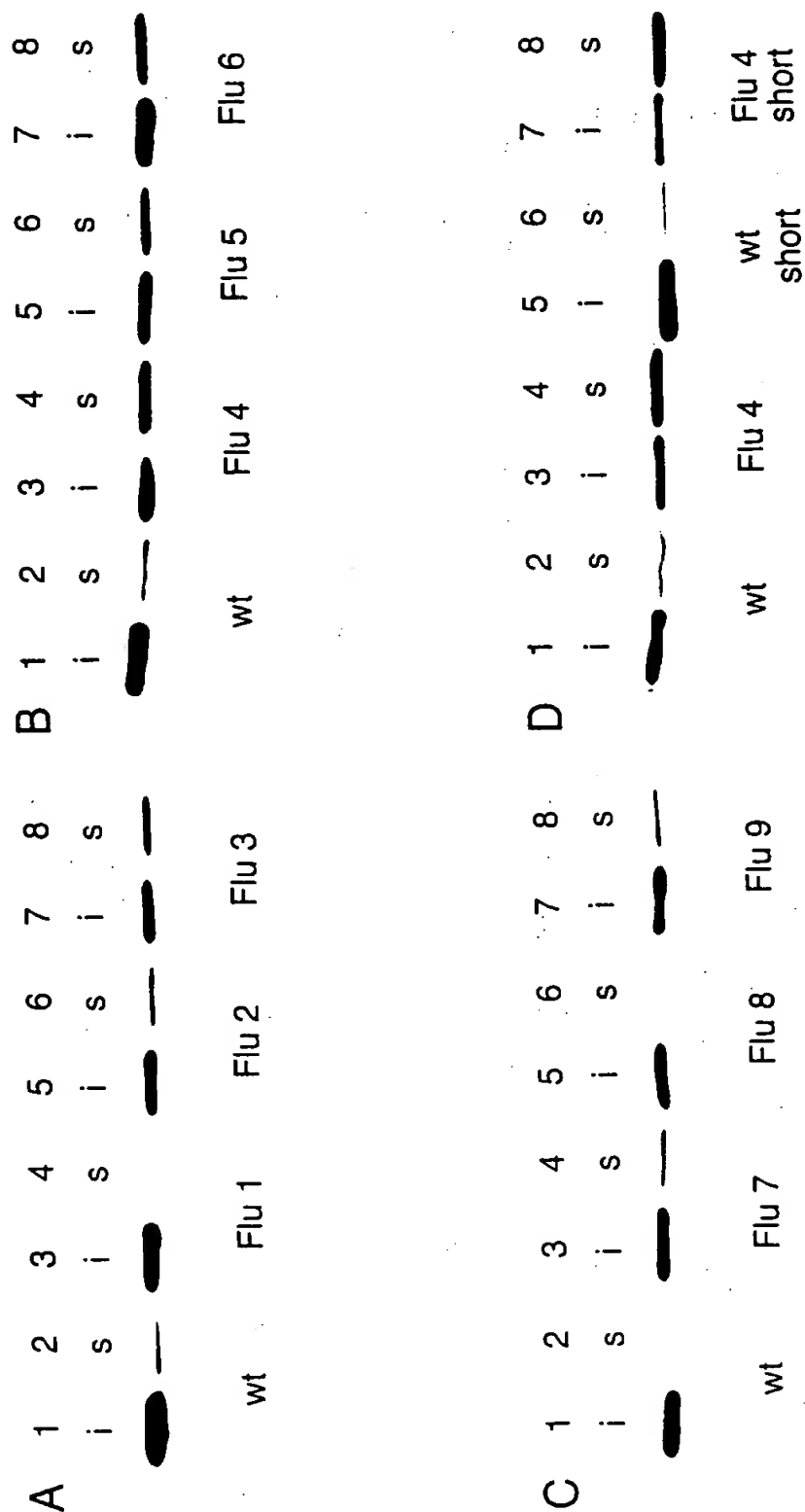


Figure 4: Scatchard plots of fluorescence titration of fluorescein with antibody: a) Titration of wt scFv; b) Titration of Flu4(V84D)

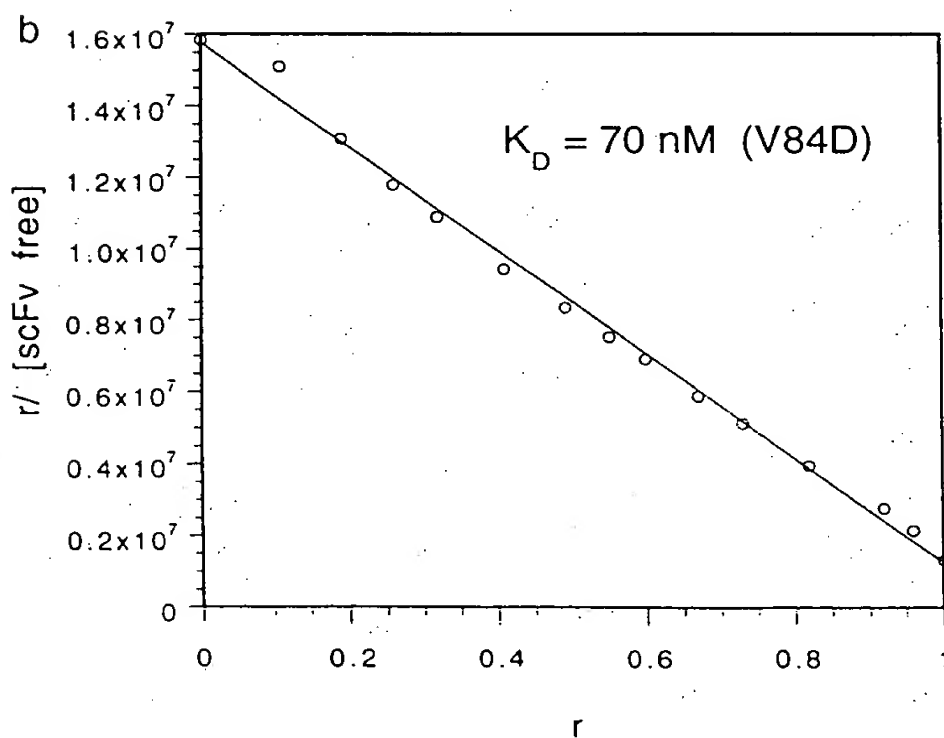
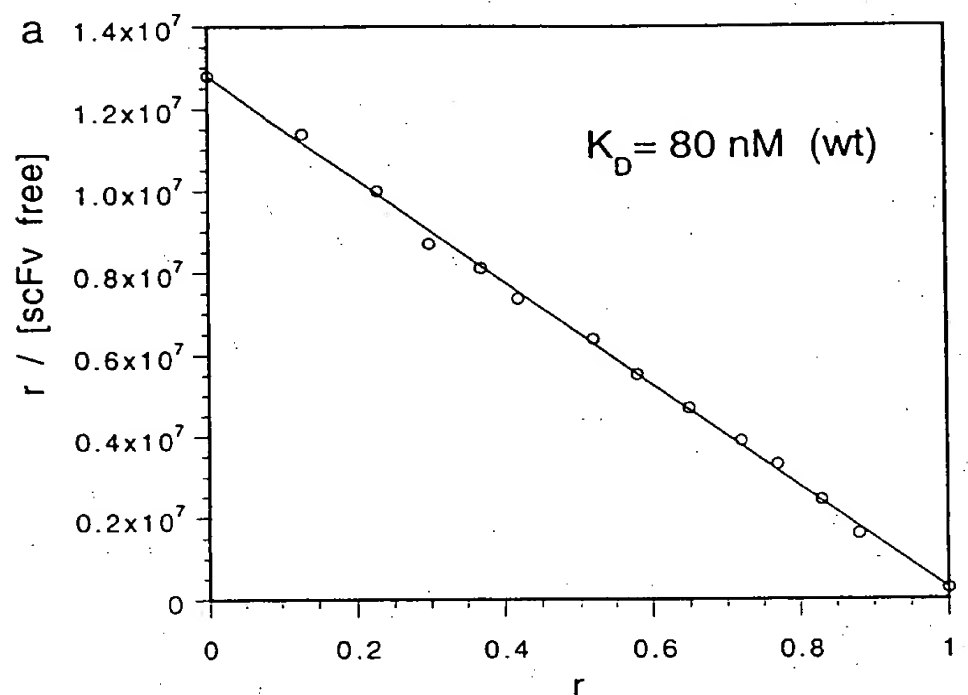


Figure 5: Overlay plot of urea denaturation. (x) wt scFv, (o) Flu4

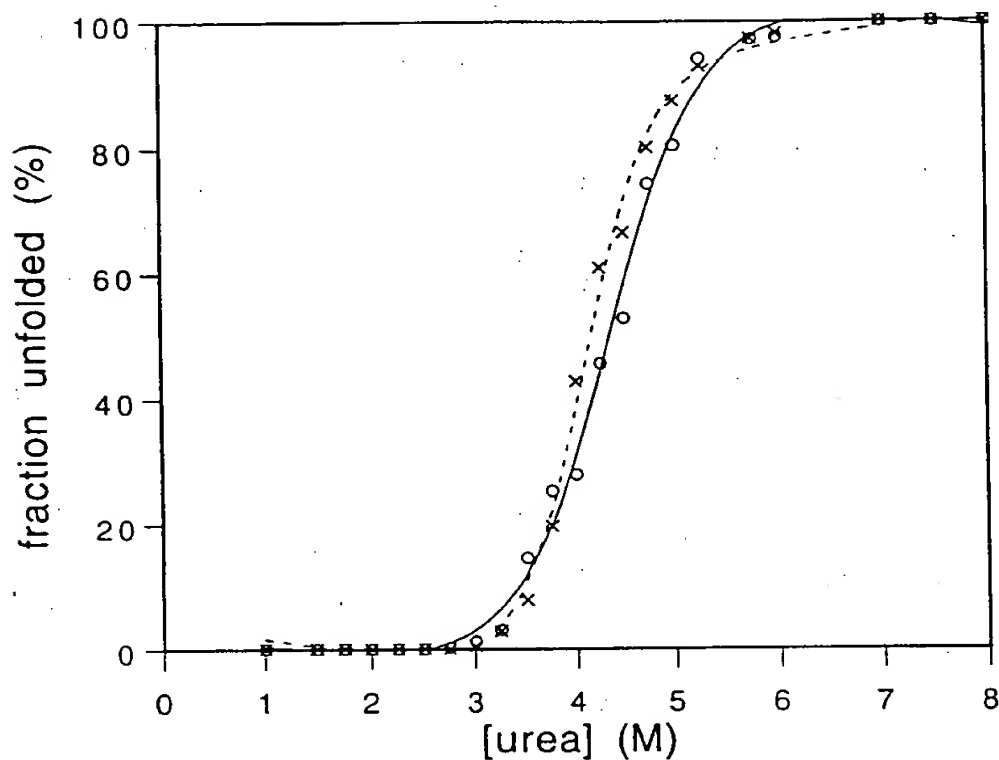
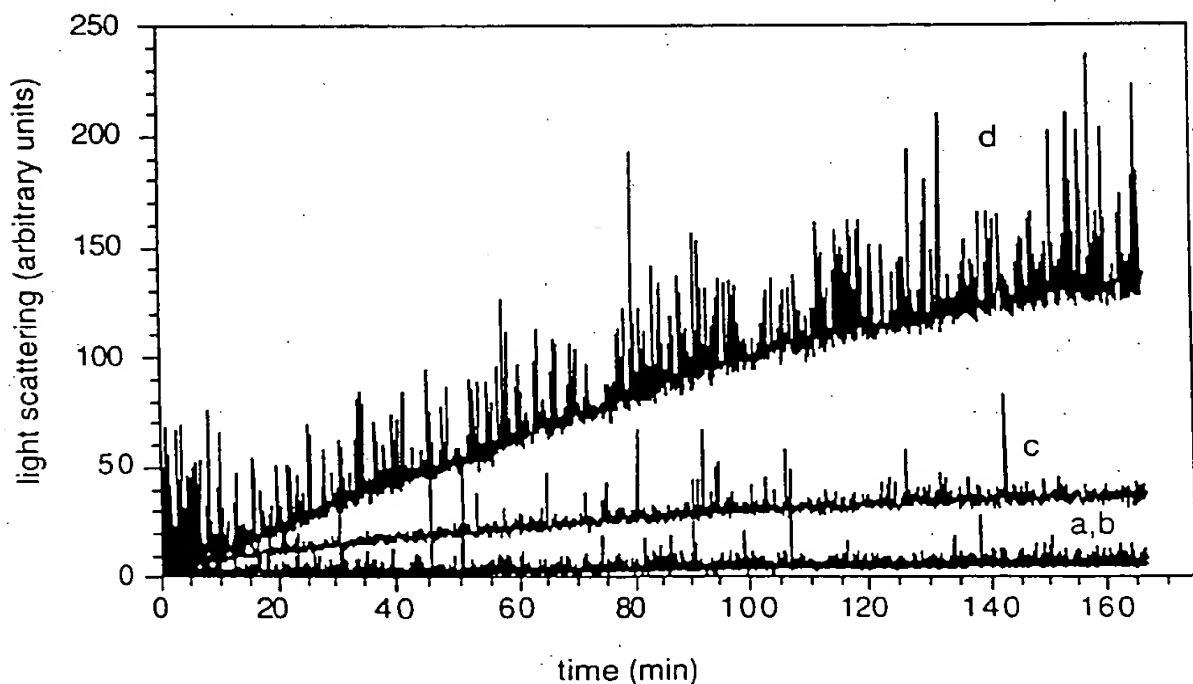


Figure 6: Thermal denaturation time courses at 40°C and 44°C for wt and Flu4 scFv fragments



A vertical strip of a musical score, likely a page from a manuscript. It features several staves with musical notation, including notes and rests. There is some text written on the page, possibly lyrics or performance instructions, interspersed with the musical staves. The strip is oriented vertically, showing a portion of the original document.

% exp. (FAB)
% exp. (ind.)
% buried v/c

Seq. 4-4-20
Consensus
Distribution:

Seq. 4-4-20
Consensus
Distribution:

Asp Glu Lys Arg His Thr Ser Asn Gln Gly Ala Cys Pro Val Ile Leu Met Phe Tyr Trp

Table 1: Sequence variability of residues contributing to the v/c interface (cont.)

L40				L41				L80				L81				L83				L103			
kappa	hu	mu	lambda	kappa	lambda		kappa	lambda		kappa	lambda		kappa	lambda		kappa	lambda		kappa	lambda			
					hu	mu		hu	mu		hu	mu		hu	mu		hu	mu		hu	mu	hu	mu
					Pro	Pro		Pro	Pro		Pro	Pro		Pro	Pro		Pro	Pro		Pro	Pro	Pro	Pro
0	0	0	0	0	10	3	96	0	0	0	0	10	7	4	0	0	0	15	0	0	0		
0	0	0	0	0	4	0	0	0	8	0	0	86	93	69	82	0	0	99	68	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	88	98		
1	0	0	0	0	1	2	0	0	0	0	0	0	0	0	0	0	0	0	0	8	1		
0	0	0	0	0	2	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
0	1	1	0	0	0	0	0	0	3	17	94	0	0	0	0	0	3	0	0	2	0		
2	19	5	0	0	1	0	0	9	6	19	0	0	0	0	0	2	0	0	0	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
0	3	0	0	0	0	0	0	0	6	0	0	0	0	1	0	0	0	0	0	0	0		
0	0	0	0	0	99	80	3	0	0	0	0	1	0	17	0	0	1	0	0	0	0		
1	1	1	1	0	0	2	0	16	64	56	4	1	0	0	0	0	31	0	0	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
94	77	92	96	0	0	0	0	74	9	2	1	0	0	0	0	0	0	0	0	0	0		
1	0	0	0	0	0	1	0	0	2	5	0	0	0	1	0	20	12	0	0	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	11	0	0	0	0		
0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	1	32	0	0	0	0		
0	0	0	3	0	0	0	0	0	0	0	0	0	0	6	0	0	2	0	0	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	72	7	0	0	0	0	0		
0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0		
0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		

Table 1: Sequence variability of residues contributing to the v/c interface (cont.)

[illegible]

Table 1: Sequence variability of residues contributing to the v/c interface (cont.)

	H9			H10			H11			H13			H14			H41			H42			H43		
	Gly	Pro	Gly	Glu	Gly	hu	mu	hu	mu	hu	mu	Pro	Pro	Pro	Ala	Arg	Pro	Pro	Pro	Gly	Gly	Lys		
Asp	0	0	5	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2		
Glu	2	0	27	54	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	1	19		
Lys	0	0	0	0	0	0	0	0	0	59	54	0	0	0	0	3	0	0	0	0	0	0		
Arg	0	0	1	0	0	0	0	0	0	3	18	0	0	0	1	36	1	0	0	0	0	1		
His	0	0	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	11	0	0	0	0		
Thr	1	2	1	0	0	0	0	0	0	0	0	1	3	1	10	13	1	1	1	0	0	0		
Ser	1	1	0	2	0	0	0	0	0	0	0	0	2	0	0	18	2	3	0	0	8	0		
Asn	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0	0	0	0		
Gln	0	0	0	0	0	0	1	0	0	34	22	0	0	0	0	0	0	0	0	0	0	76		
Gly	42	29	61	38	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	90	2		
Ala	33	31	3	1	0	0	0	0	0	0	5	8	1	0	59	7	1	0	0	0	1	2		
Cys	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
Pro	21	36	0	0	0	0	0	0	0	96	85	0	0	0	18	14	94	83	0	1	0	0		
Val	0	1	0	1	0	38	1	0	0	0	0	0	0	0	1	1	0	0	0	0	0	0		
Ile	0	0	0	0	0	0	2	0	0	0	0	3	0	0	1	1	0	0	0	0	0	0		
Leu	0	0	0	0	0	60	95	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0		
Met	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0	0		
Phe	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
Tyr	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
Trp	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		

Position
% exp. (FAB)
% exp. (ind.)
% buried v/c
Species
Seq. 4-4-20
Consensus
Distribution:

Table 1: Sequence variability of residues contributing to the v/c interface (cont.)

H84			H87			H89			H105			H108			H110			H112			H113		
Ala	Ser	Val	Thr	Ser	Met	Val	Val	Ile	Gln			Ser			Thr	Thr	Ser	Ser	Ser	Ser	Ser		
									hu	mu	hu	mu	hu	mu								hu	mu
1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	72		
0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	78		
0	0	0	0	0	0	0	0	0	10	0	0	0	0	0	0	0	0	0	0	0	7		
0	0	0	0	0	0	0	0	1	5	0	0	0	0	0	0	0	0	0	0	0			
0	0	0	0	0	0	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0			
4	12	0	96	46	0	4	15	0	0	4	0	23	51	89	99	0	98	100	0	97	77		
18	70	3	2	51	0	0	0	0	0	0	0	0	23	2	0	0	0	0	0	0	0		
0	0	0	0	0	0	0	0	0	80	0	83	0	0	0	0	0	0	0	0	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1		
55	12	0	0	1	0	0	0	0	0	11	0	0	0	0	0	0	0	0	0	0	22		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
14	1	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	1	0	0	2	0		
4	0	1	0	0	0	76	50	0	0	0	0	0	0	1	0	0	1	0	0	0	0		
1	0	1	0	0	0	5	13	0	0	0	0	0	0	7	0	0	0	0	0	0	0		
0	0	0	0	0	0	6	7	0	0	0	0	63	25	0	0	0	0	0	0	0	0		
0	0	0	0	1	0	7	13	0	0	0	0	9	0	0	0	0	0	0	0	0	0		
2	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		

Table 2: Mutations introduced in the scFv fragment of the antibody 4-4-20

	L15E (VL)	L11N (VH)	L11D (VH)	V84D (VH)
Flu 1	•			
Flu 2		•		
Flu 3			•	
Flu 4				•
Flu 5		•		•
Flu 6			•	•
Flu 7	•	•		
Flu 8	•		•	
Flu 9	•	•		•
Flu 4 short				•

Table 3: K_D values of the different scFv mutants determined in fluorescence titration

	Flu wt	Flu 3	Flu 4	Flu 6	Flu wt#
K_D (nM)	80 ± 7	60 ± 12	70 ± 10	75 ± 13	90